Associating MRI Findings with Molecular Profiles Using Image-Guided Genomic and Proteomic Microarray Analysis

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To understand the molecular pathophysiology of cancer, genomic expression profiles of tumors are being correlated with clinical presentation patterns, surrogate disease markers, and pathological evaluations.[1-9] Solid tumors are, however, spatially and temporally heterogeneous and display different morphologies within the same tumor and at each stage of disease progression.[10-12] Magnetic resonance imaging (MRI) is a powerful clinical tool with high spatial and temporal resolution that can discern different regions within a contiguous tumor for biopsy and tissue analysis.[13,14]

In this presentation, we will discuss the concepts in state of the art genomic and proteomic microarray analysis as it relates to cancer research. For genomic analysis we will discuss spotted and oligonucleotide microarray techniques. The advantages and disadvantages of each approach, and bioinformatic updates in analyzing the large array of information resulting from this high throughput technique will be evaluated. The role of imaging and in particular MRI-guided tissue sampling will be discussed. Similarly, current proteomic techniques and parallel functional advantages of this technique will be discussed. Results from MRI-guided sampling of tissue for microarray analysis will be presented to show the power of this approach in obtaining the most relevant therapeutic targets.

The future of radiologic imaging and pathologic evaluation of cancer is moving from gross visualization of shapes to molecular and functional profiles. With the completion of the human genome project and development of high throughput microarray analysis, we are now able to obtain the state of the neoplasm at the molecular level at the time of tissue sampling. Through classification of patients with similar molecular profiles, it is now evident that finer classifications of the neoplasms exist than can not be visualized classically under the scope of the pathologist. Solid tumors are heterogeneous, and the ability to determine patients that will respond to certain chemotherapeutic regiments or improved prognostic indicators will have a significant impact in the practice of medical oncology. Since ~34,000 genes can now be evaluated with microarray techniques, the expression variability from patient to patient, spatial and temporal heterogeneity, and the general variablitiy introduced in the experimental processing makes the data very noisy and finding relevant information very difficult. We propose that image-guided MRI is a powerful functional imaging technique that will facilitate the extraction of the most relevant targets for targeted imaging and therapeutic development.

The temporal changes as a small tumor progresses towards a large tumor with necrotic centers is easily reflected in the contrast enhanced T1 and T2- weighted images. As

demonstrated from the series of images in Figure 1, at an early stage of tumor growth, the tumor is homogeneous in both T1-contrast and T2-weighted images, H&E stained tissue samples, and molecular profiles. However, at an intermediate stage of tumor growth, differential contrast uptake highlights areas of viable tumor with little to no vascular permeability and areas of viable tumor with high vascular permeability to contrast agents. This is not discernable in H&E staining but is reflected in genomic and proteomic profiles. This is an important stage of tumor growth as most patients become symptomatic at this or later stages. Finally, tumors can progress to develop necrotic centers that have radically different T1 and T2 characteristics, H&E and microarray profiles.

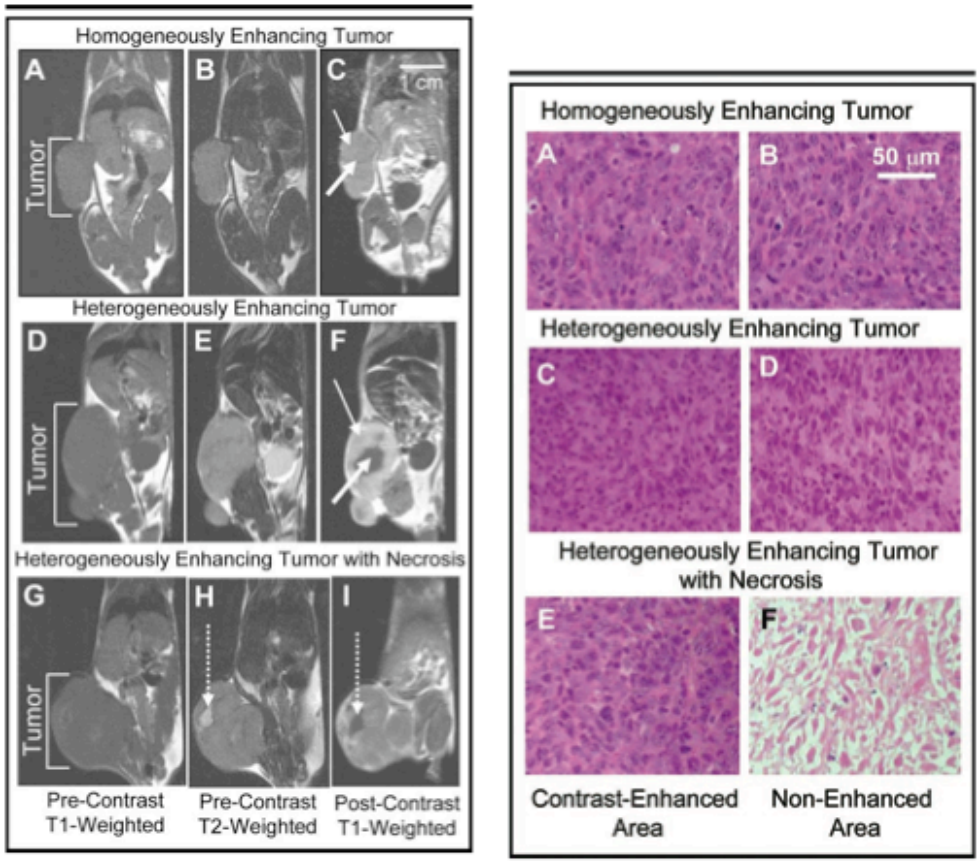

Figure 1. Temporal changes of tumor progression as correlated with H&E staining[15,16].

The genomic and proteomic profile for the heterogeneously enhancing tumors prior to necrosis indicate significant extracellular matrix remodeling in the contrast-enhancing areas as compared to the non-enhancing areas. This is in agreement with the hypothesis that the contrast-enhancing region is the active, aggressive area of tumor growth where angiogenesis and metastisis occur. If patients present with tumors at this stage of growth, treatment regimens that include antiangiogenic approaches may have significant impact. The molecular profile for tumors with necrotic regions show upregulation of genes

related to leukocyte infiltration around the rim of necrosis. Patients presenting with these necrotic centers may benefit from immunotherapy.

The First clinical application of MRI guided tissue sampling for primary brain tumors will be presented. The use of the targets identified using this method for molecular imaging and antiangiogenic approaches will be presented. This technique is currently in preparation for phase one clinical trials.

It is clear that we have now only begun effective sampling, and mining of data generated by high throughput microarray systems and the impact of this is evident in drug development by the increasing number of drugs in clinical trials that target these relevant molecular pathways (Avastin, Erbitox, ….). The advances in functional MRI techniques provide a unique, high resolution method to distinguish characteristics of hetergenous regions of the tumor, understand the pathophysiology, design new drugs, and finally develop effective therapeutic plans for patients.

References

1. Bittner, M. et al. Molecular classification of cutaneous malignant melanoma by gene expression profiling. *Nature* **406**, 536-540 (2000).

2. Perou, C. M. et al. Molecular portraits of human breast tumours. *Nature* **406**, 747-752 (2000).

3. Alizadeh, A. A. et al. Distinct types of diffuse large B-cell lynphoma identified by gene expression profiling. *Nature* **403**, 503-511 (2000).

4. Golub, T. R. et al. Molecular Classification of Cancer: Class Discovery and Class Prediction by Gene Expression Monitoring. *Science* **286**, 531-537 (1999).

5. Duggan, D. J., Bittner, M., Chen, Y., Meltzer, P. & Trent, J. M. Expression Profiling Using cDNA Microarrays. *Nature Genetics* **21**, 10-14 (1999).

6. Pinkel, D. Cancer cells, chemotherapy and gene clusters. *Nature Genetics* **24**, 208-209 (2000).

7. Scherf, U. et al. A gene expression database for the molecular pharmacology of cancer. *Nature Genetics* **24**, 236-244 (2000).

8. Khan, J. et al. Gene expression profiling of Alveolar Rhabdomyosarcoma with cDNA microarrays. *Cancer Research* **58**, 5009-5013 (1998).

9. Coller, H. A. et al. Expression analysis with oligonucleotide microarrays revels that MYC regulates genes involved in growth, cell cycle, signaling, and adhision. *Proc. Natl. Acad. Sci. USA* **97**, 3260-3265 (2000).

10. Ross, J. S., Sheehan, C. E., Dolen, E. M. & Kallakury, B. V. S. Morphologic and molecular prognostic markers in prostate cancer. *Advances in Anatomic Pathology* **9**, 115-128 (2002).

11. Wilkinson, N. & Rollason, T. P. Recent advances in the pathology of smooth muscle tumours of the uterus. *Histopathology* **39**, 331-341 (2001).

12. Kuhl, C. K. & Schild, H. H. Dynamic image interpretation of MRI of the breast. *Journal of Magnetic Resonance Imaging* **12**, 965-974 (2000).

13. Taylor, J. S. et al. MR Imaging of Tumor Microcirculation: Promise for the New Millenium. *Magnetic Renosance Imaging* **10**, 903-907 (1999).

14. Yuh, W. T. C. An Exciting and Challenging Role for the Advanced Contrast MR Imaging. *J. Magn. Reson. Imaging* **10**, 221-222 (1999).

15. Guccione,S. et al. Functional Genomics as Guided by MRI Imaging: Mouse Tumor Model Study. *Radiology* **228**, 560–568 (2003).

16. Ying, Y., Guccione, S., Bednarski, M. Comparing Genomic and Histologic Correlations to Radiographic Changes in Tumors: A Murine SCC VII Model Study. *Academic Radiology*,**10**, 1165–1175 (2003).